Mechanism of Migration from Agglomerated Cork Stoppers: I. An Electron Spin Resonance Investigation

THÉRÈSE SIX,[1] ALEXANDRE FEIGENBAUM,[1] ANNE-MARIE RIQUET[2]

[1] Institut National de la Recherche Agronomique, Moulin de la Housse 51687 Reims, France

[2] Institut National de la Recherche Agronomique–LNSA, Domaine vilvert, 78350 Jouy-en-Josas, France

Received 13 November 2000; accepted 6 July 2001

ABSTRACT: Related to the study of interactions between food and packaging, migration from agglomerated cork stoppers was studied by electron spin resonance. Paramagnetic probes were incorporated separately in the adhesive or in the cork granules. The finished cork was obtained by an individual molding procedure. The behavior of aminoxyl probes, differing by their functional groups, was studied. Free probes, initially incorporated into the adhesive phase, partitioned into cork during processing. Their migration occurred from both the cork and the adhesive phases. With amino-TEMPO, a probe covalently bonded to the adhesive phase, it was possible to study the penetration of the alcoholic simulant of wine (12% ethanol) into the whole structure of the cork and to demonstrate that strong swelling took place. © 2002 John Wiley & Sons, Inc. J Appl Polym Sci 83: 2644–2654, 2002

Key words: cork; adhesives; polyurethanes; ESR; migration

INTRODUCTION

Agglomerated stoppers are composed of natural cork granules (containing mainly suberine, lignine, and polysaccharides) and adhesives. The latter are usually polyurethanes with reactive isocyanide end groups, which react with the moisture present in cork to give chain coupling and crosslinking during processing.[1] As far as food safety aspects are concerned, one has to take into account the behavior of residual precursors (usually toluene diisocyanide or methylene bisisocyanide) and the possible low-molecular-weight transformation products (the corresponding ureylenes and aromatic amines). Additives may also migrate into wine. For this reason, it is often recommended that the composition of these adhesives be approved by regulatory bodies.[2]

Pending the development of such regulations, scientific knowledge on the mechanism of migration from cork must be obtained. Typically, a cork (10 g) contains 0.8 g of adhesive. The mass of adhesive used for stopping 1 L of wine is thus similar to the mass of coating in a 1-kg food can. Can coatings were recently shown to release significant amounts of migrants to foodstuffs, which justifies a specific regulation.[3] Obviously, a major difference between cork adhesives and coatings lies in the surface in contact with the food product. The surface of adhesive in direct contact with wine is negligible. However, several mechanisms could contribute to a significant migration of adhesive constituents. First, if water penetrates into the cork, it could significantly increase the surface of contact. Second, if the potential migrants diffuse from the adhesive to the cork, the whole cork could become a reservoir of migrants, and the migration into wine could be greatly facilitated. It was one of the purpose of this work to investigate whether the latter mechanisms occur.

Correspondence to: A.-M. Riquet.

Journal of Applied Polymer Science, Vol. 83, 2644–2654 (2002)

DOI 10.1002/app.10230

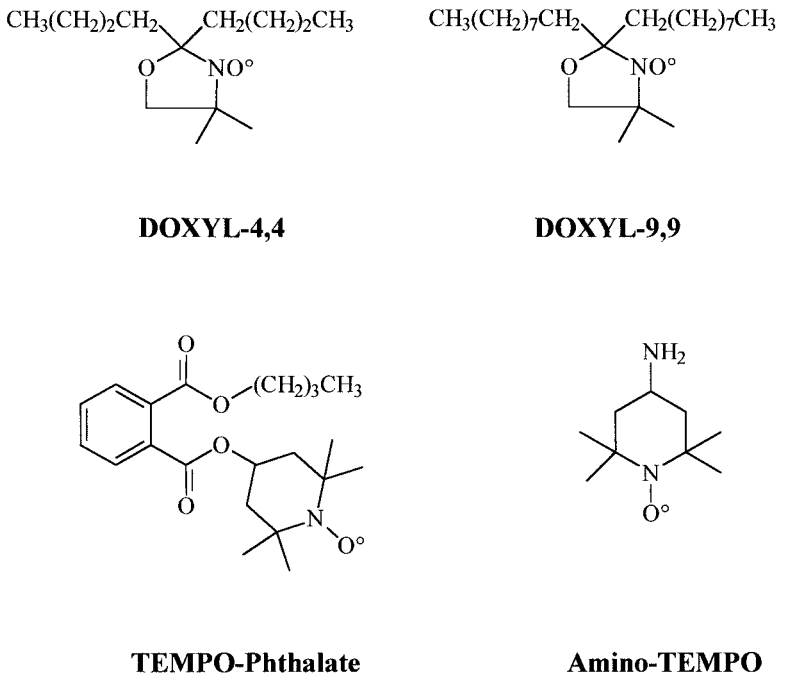

Figure 1 Paramagnetic probes.

Food and packaging interactions are usually demonstrated in an indirect way by monitoring the migration of chemicals released to food or food simulants. The penetration of food (or food simulants) into the material has been studied by monitoring of the weight uptake of the materials in function of time,[4] by ultraviolet or IR spectroscopy,[5,6] by magnetic resonance imaging,[7] and with radioactive[8] or paramagnetic probes.[9,10]

In the first part of this study, we selected the latter approach. Paramagnetic probes are usually detected selectively in materials by electron spin resonance (ESR), and the analysis of line shapes and their temperature dependence provides useful information on the polymeric network.[11] This approach recently proved to be very powerful for the study of the interaction of food simulants with multiphasic polymers.[12]

EXPERIMENTAL

Paramagnetic Probes

The paramagnetic probes are described in Figure 1. Two types of probes were selected, according to their reactivity.

Nonreactive probes included TEMPO-phthalate, which has a structure close to that of bis(2-ethylhexyl) phthalate, a classical plasticizer of adhesives. DOXYL-4,4 and DOXYL-9,9 are linear probes, which according to the number of carbons in the chain, may behave differently in the polymeric network.

The reactive probe was amino TEMPO, which due to its amino group, may graft on adhesive polymers, especially by reaction with isocyanide groups.

TEMPO-phthalate and DOXYL-4,4 were synthesized in high yield in the laboratory.[13] DOXYL-9,9 and amino-TEMPO (AT) were commercial products available from Aldrich Chemical (Saint-Quentin Fallavier, France).

Labeling of Each Constituent of the Finished Cork Separately

Labeling of the Adhesive

The adhesive studied is a polyurethane prepolymer with isocyanide chain ends (Fig. 2; NOVAFLEX NM15 ATO Findley). These functions react with atmospheric moisture and with that of the cork granules.

DOXYL-4,4 (1200 ppm) or DOXYL-9,9 (343 ppm) were added to the prepolymer (non-polymerized adhesive) at room temperature in a mixer (350 rpm for 5 min). Because TEMPO-phthalate is solid at room temperature, blending (1500 ppm) was realized at 60°C.

We grafted AT by mixing 1400 ppm (5.6 mg) of probe with the adhesive (4 g) in an ultrasonic bath for 1 h at 60°C.

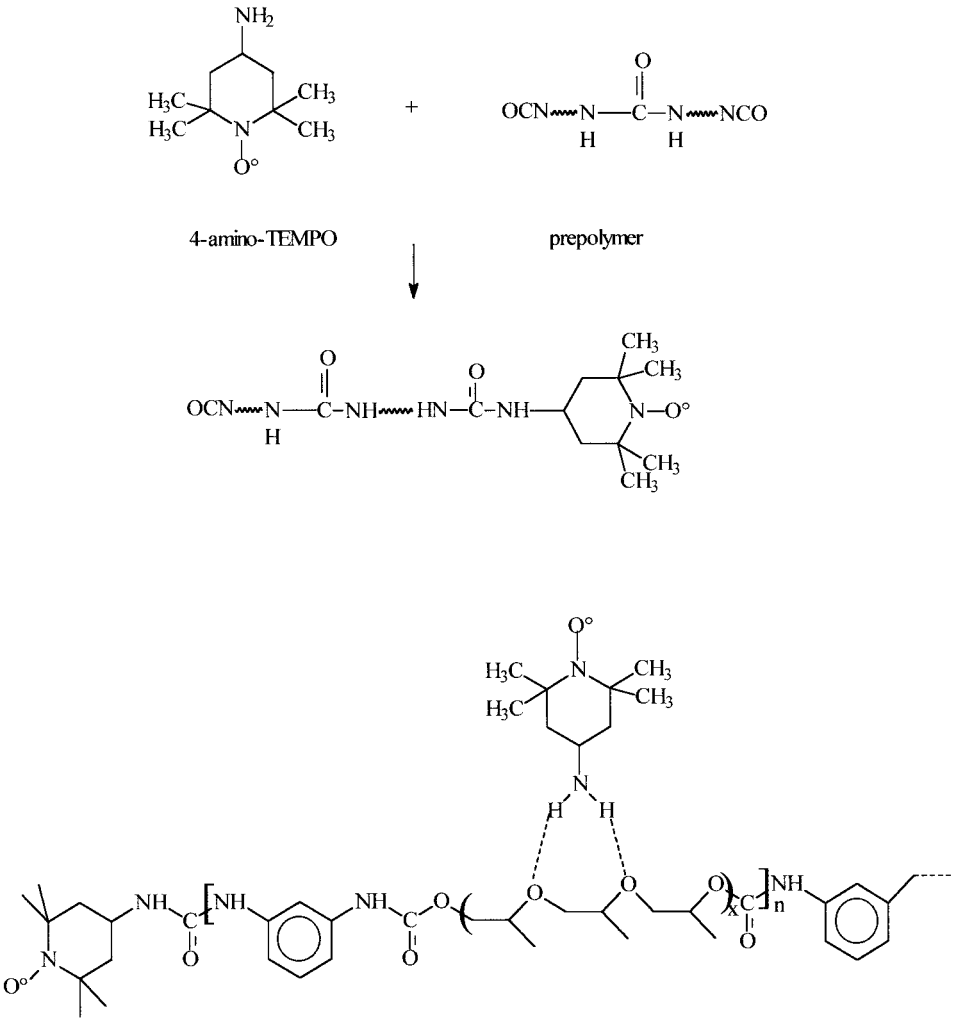

Figure 2 Reaction of AT with adhesive.

A film of labeled, polymerized adhesive was obtained by exposure of the prepolymer to atmospheric moisture (98% relative humidity) at room temperature for 1 day.

Labeling of Cork Granules

To allow diffusion and homogenous distribution of nonreactive probes in cork granules, the cork granules (4 × 20.75 mg) and the probe (two drops) were placed in a hermetic poll box kept in an oven at 40°C for 30 min (DOXYL) and at 70°C for 2 h (TEMPO-phthalate). The probe, thus, evaporated and diffused into the granules.

Processing of Finished Corks

The finished cork was obtained by an individual molding procedure of the mixture [10 g of cork granules + 1.75 g of adhesive (extreme quantity) + 0.3 g of Vaseline + probe]. Vaseline facilitated the removal of the processed cork from the mold.

The mixture was prepared in a reactor and stirred for 15 min. Then, it was put in a mold, pressed, and heated at 125°C for 45 min. The final dimensions were obtained by turning and sandpapering (diameter = 31 mm, height = 34 mm).

Conditions of Contact with the Simulant

To study the possible alterations of finished cork related to contact with the alcoholic simulant, we immersed the labeled corks in 250 mL of ethanol (12% solution in distilled water) at pH 3 (adjusted with citric acid) for 10 days at 40°C. Corks were then cut lengthwise in thin slices (1 mm thick), dried at 30°C (8 h), weighed, and placed into ESR tubes before being analyzed. A nonimmersed sample was used as a reference.

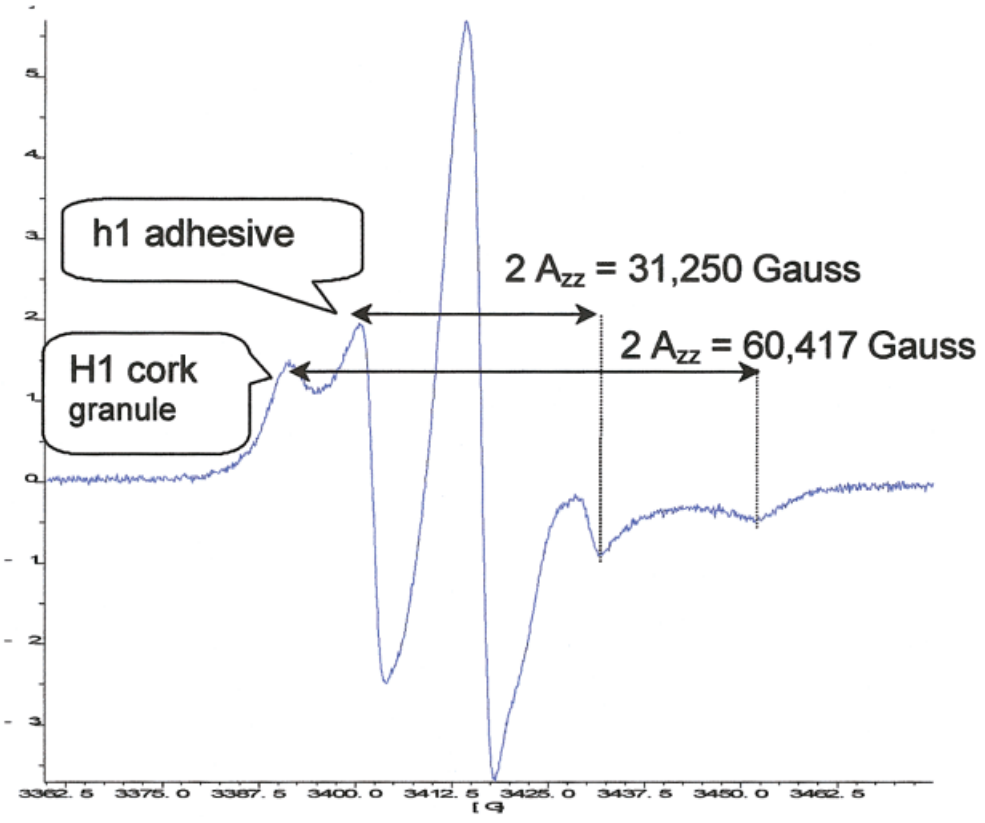

Figure 3 ESR spectrum of stopper containing DOXYL-4,4 or DOXYL-9,9.

Study by ESR

ESR spectra were measured with a Bruker ESP 300 spectrometer (Wissembourg, France) in the temperature range −50 to 150°C. The weighed samples (3 × 1 × 20 mm slices) were placed in an ESR tube (diameter = 32 mm, height = 18 cm) and analyzed directly in the spectrometer cavity. To evaluate migration, we assessed the proportion of the probe in the material from the area (double integration) of the signal per mass unit. To evaluate partition of the probe between different environments, we measured the ratio of specific lines h_{-1} and H_{-1} (Fig. 3).

The hyperfine split ($2A_{zz}$) of the spectrum was measured as the distance (in Gauss) between the two extreme peaks. The temperature of fast signal (t_{fs}) corresponds to the particular temperature at which the fast signal was differentiated from that of the immobilized signal on the spectrum. T_{50G}, the temperature characteristic of the mobility of the probe, was defined experimentally[11] as the temperature where the width of the signal $2A_{zz}$ is equal to 50 Gauss. The lower T_{50G} is, the higher the mobility of the radical is.

Identical spectra were always obtained by application of the same temperature sequence a second time. Values of T_{50G} were measured both before and after contact with the simulant.

Differential Scanning Calorimetry (DSC) Measurements

DSC measurements were made with a TA Instruments DSC 2920 (Saint-Quentin en Yveline, France).

Samples of adhesive [before (3.9 mg) and after (4.1 mg) contact with the simulant] were initially heated from −100°C (5 min) to 110°C (10 min) at a heating rate of 10°C/min. Glass-transition temperatures were measured by the midpoint method.

RESULTS AND DISCUSSIONS

Agglomerated cork stoppers are layered by a silicone and/or paraffin film, and the side that is toward the wine is generally covered by a disk of natural cork. However, in this study, we worked only on the agglomerated part and call it *finished cork* by definition.

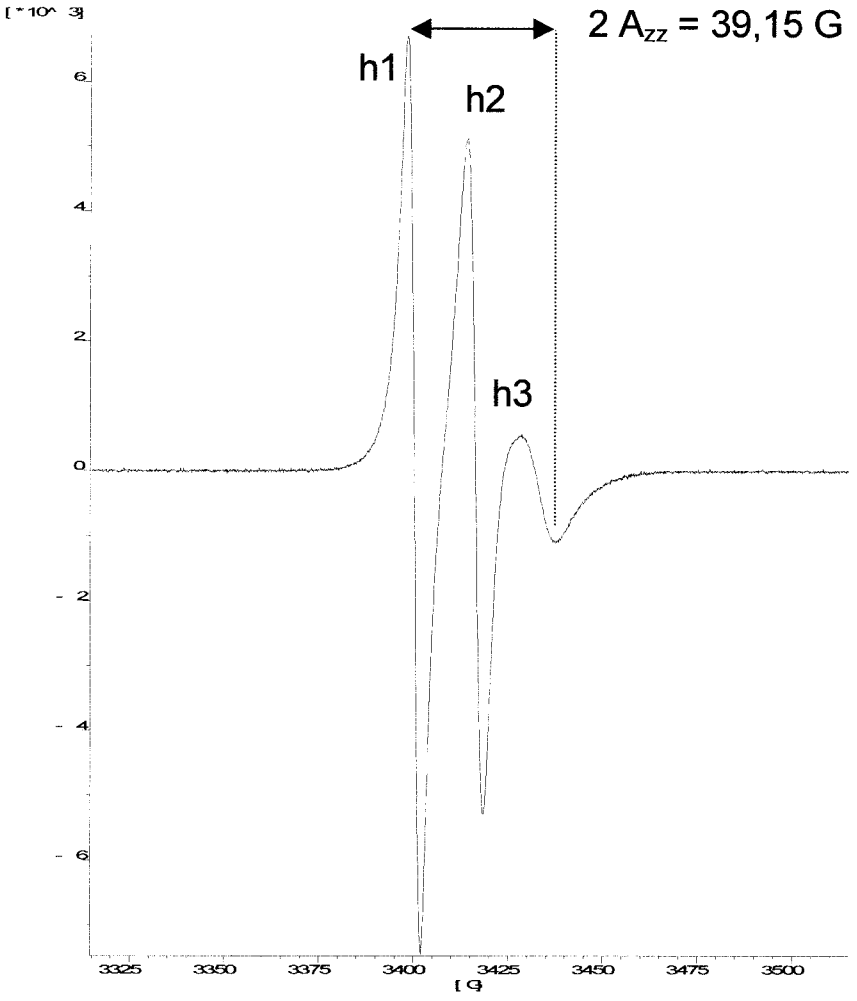

Figure 4 ESR spectrum of TEMPO-phthalate in the nonpolymerized adhesive.

Each probe was incorporated separately in the materials studied. The following probes were selected for the investigation on cork and cork adhesives:

- DOXYL-4,4, DOXYL-9,9, and TEMPO-phthalate were initially dispersed in the adhesive and could diffuse freely into the cork.
- AT is a reactive molecule, which may graft on the polymeric chain by reaction with isocyanides. This probe cannot diffuse into the cork but behaves like a camera bound to the adhesive. The TEMPO group is necessarily attached to chain ends of the polymer[12] (Fig. 2).

Behavior of Probes Dispersed in Cork, Adhesive, and the Finished Cork

Before investigation of probes in the finished cork, we studied their behavior separately in cork and in adhesive.

Probes Dispersed in the Adhesive

The ESR signal of TEMPO-phthalate in the nonpolymerized adhesive ($2A_{zz}$ = 39.15 Gauss) is shown in Figure 4. It consisted of three sharp lines (at room temperature), which are characteristic of a probe that can move freely ($5.10^{-11} < \tau_r < 10^{-9}$ s) relative to the direction of the magnetic field of the spectrometer.[11]

When the adhesive was polymerized by simple exposure to atmospheric moisture (98% relative humidity) for 1 day, the spectrum of the probe, recorded at room temperature, was very close to that shown in Figure 4.

The other dispersed probes, DOXYL-4,4 and DOXYL-9,9, incorporated separately in the adhesive, gave similar spectra.

Probes Dispersed in Cork Granules

The ESR spectrum of TEMPO-phthalate in cork is shown in Figure 5. Such a broad spectrum ($2A_{zz}$

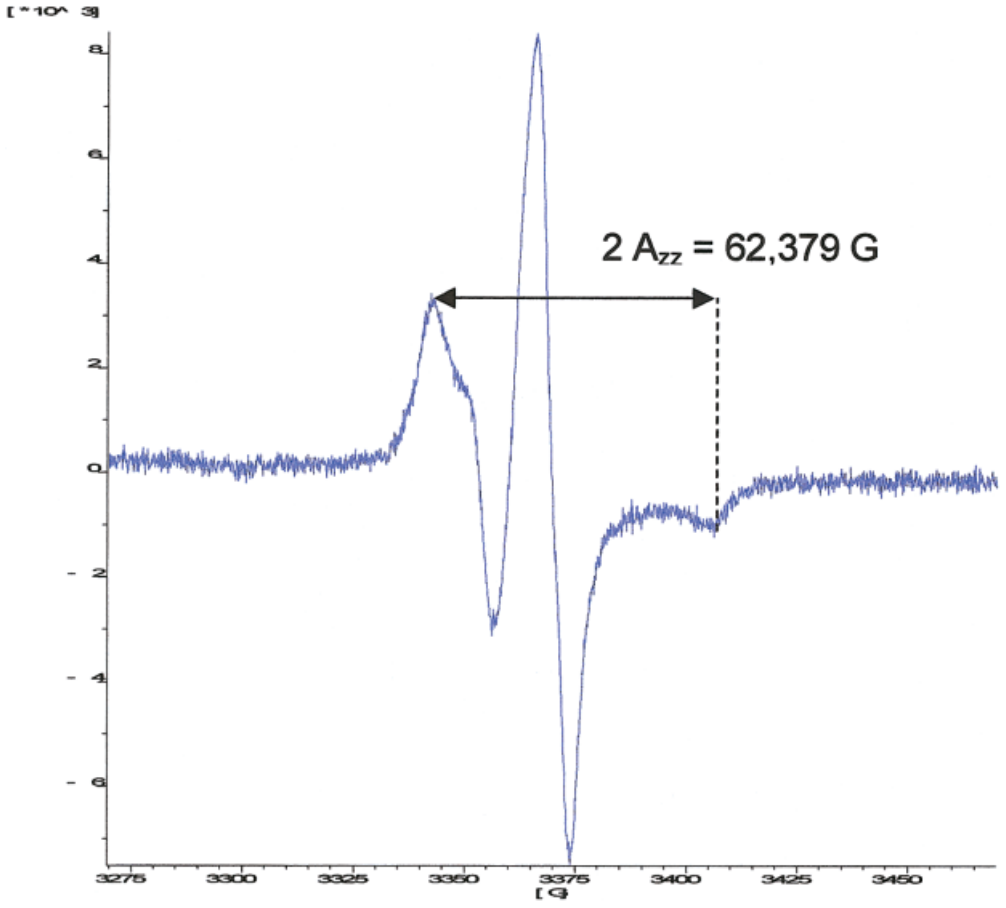

Figure 5 ESR spectrum of TEMPO-phthalate in cork granules.

= 62.37 Gauss) is characteristic of a probe whose movements relative to the magnetic field are slow ($5.10^{-9} < \tau_r < 10^{-7}$ s) in the cork matrix. We write here of a "hindered probe." The shape of the spectra were similar with DOXYL ($2A_{zz}$ = 60.4 Gauss).

The different behavior of the probes in cork and in pure adhesive suggested that it would be possible to differentiate the two phases in a finished cork.

Probes Dispersed in Finished Corks

Adhesive doped with probes were thoroughly mixed with cork granules; stoppers were then processed at 125°C, in conditions replicating those on a plant. The spectrum of a stopper containing DOXYL-4,4 or DOXYL-9,9 (Fig. 3) clearly displays the superimposition of the two spectra shown in Figures 4 and 5. The free probe ($2A_{zz}$ = 31.25 Gauss, adhesive phase) was well differentiated from the hindered probe ($2A_{zz}$ = 60.417 Gauss, cork phase). By comparing the spectra in adhesive and in cork granules, one can attribute the two spectra to two distinct environments: the free probe in the adhesive phase and the hindered probe in the cork phase.

With TEMPO-phthalate at room temperature, the hindered probe (characteristic of the cork phase) was only detected as a shoulder (Fig. 6). To fully confirm this allocation, we recorded the spectrum of a finished cork sample containing TEMPO-phthalate at a lower temperature, as it is well known that line shapes are sensitive to temperature. Indeed, at −20°C (the rigid limit spectrum of the probe in cork), the characteristic superimposition of the two probe environments was observed.

These results show clearly that the nonreactive probes, initially incorporated into the adhesive, partitioned into the cork phase during processing. A partition coefficient could be estimated by mathematical modelization of the spectra. However, although a precise value of the partition coefficient was not necessary, a rough estimation of the partition was obtained from the ratio of line heights (h_{-1}/H_{-1}). These partition ratios (cork/

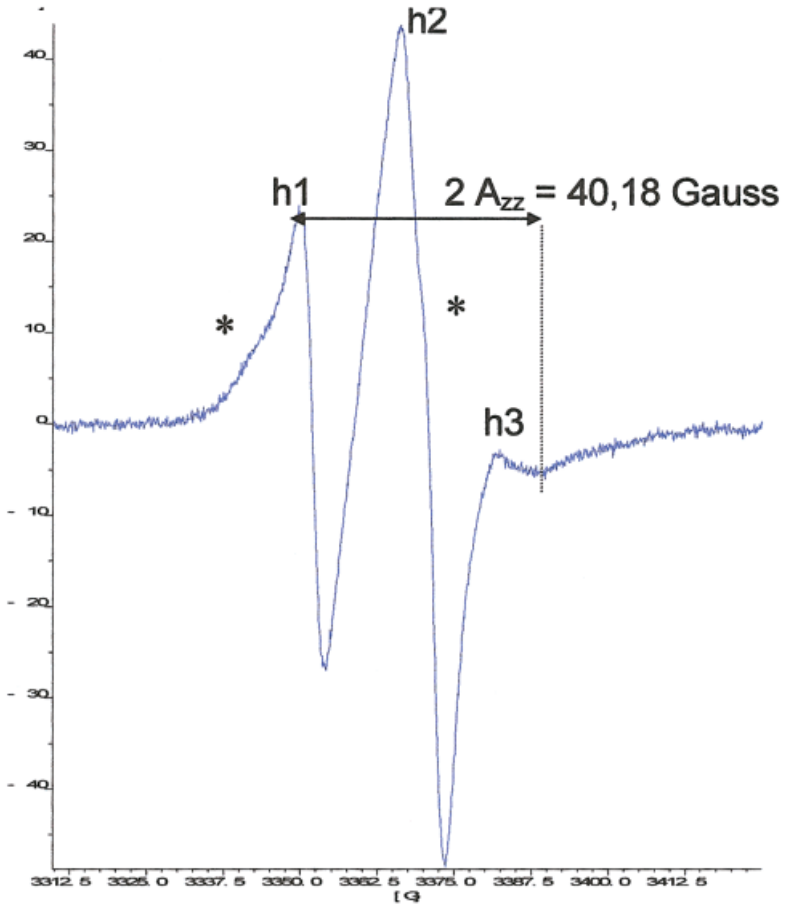

Figure 6 ESR spectrum of stopper containing TEMPO-phthalate. Shoulders correspond to the hindered probe in cork phase.

adhesive) are given in Table I. They tended to follow the polarity of the probes.

If some probes can partition between adhesive and cork phases, any other organic molecule can do the same. This means that the stopper behaves as a reservoir of some potential migrants.

Behavior of Dispersed Probes in Materials After Contact with Wine Simulant

A film of adhesive containing the TEMPO-phthalate probe was put in contact with simulated wine for 10 days at 40°C; the ESR spectrum corresponded to a very weak signal, indicating that an almost complete migration into the liquid had occurred. We tried to investigate what had happened with the probe in the cork phase of the stopper after the same treatment by recording the spectrum of stopper slices at different temperatures. Due to the weak signal and the bad differentiation between the two spectra (adhesive and cork phases), this was not very easy. We, therefore, repeated the study with stoppers containing DOXYL-4,4, which partitioned much greater than TEMPO-phthalate into the cork.

The results gathered in Table II show that the h_{-1}/H_{-1} ratio did not change significantly during contact, although the overall signal intensity decreased. This shows that migration occurred significantly from the stopper in about the same

Table I Behavior of Probes Dispersed in Finished Corks

Probe	M_w	Volume (Å^3)	h_{-1}/H_{-1}	μ(D)
TEMPO-phthalate	376		> 5	3.210
DOXYL-4,4	236	244	1.2[a]	2.096
DOXYL-9,9	326	413	0.7[a]	2.096

D = Debyes. M_w = weight-average molecular weight.
[a] Estimated close to the surface, at 0.5 cm in depth and in the middle of the cork along the axis, $n = 3$.

Table II Losses of Probes in Finished Cork After Immersion in 12% Ethanol

	Standard Kept 10 Days at 40°C		Sample After Contact 10 Days at 40°C in 12% EtOH		Migration (%)
	A/mg	h_{-1}/H_{-1}	A/mg	h_{-1}/H_{-1}	
DOXYL-4,4					
Surface[a]	0.207 ± 0.039	1.0 ± 0.2	0.159 ± 0.023	0.9 ± 0.3	23
Surface + 5 mm[a]	0.218 ± 0.043	1.0 ± 0.2	0.185 ± 0.040	0.9 ± 0.2	15
Middle[a]	0.249 ± 0.009	0.9 ± 0.1	0.214 ± 0.019	0.8 ± 0.3	14
TEMPO-phthalate					
Surface[a]	0.048 ± 0.011		0.023 ± 0.004		52
Surface + 5 mm[a]	0.071 ± 0.021		0.035 ± 0.009		51
Middle[a]	0.086 ± 0.028		0.044 ± 0.005		49

A/mg = Area of the spectrum/weight of the sample in mg.
[a] Average of eight samples.

ratio from the cork and from the adhesive phases or that both phases were in fast equilibrium.

We determined concentration gradients of the probe by cutting slices close to the surface, at 0.5 cm in depth and in the middle of the stopper along the axis. Before contact, there was a homogeneous distribution of the probe DOXYL-4,4 all over the stopper. With TEMPO-phthalate, there seemed to be a higher concentration in the middle of the stopper, which is yet unexplained (Table II). Although this difference was not far from the experimental error, it could have been due to the process.

After contact, it could be seen (Table II) that migration occurred mainly from the surface, as migration was slightly more important there (23%) than in the center (14%). Migration of DOXYL-4,4 was much less important than that of TEMPO-phthalate.

The important migration from the center of the cork indicated either a quick diffusion of the probe into the stopper or a strong penetration of the simulant into the whole structure of the stopper (this problem is rediscussed later). Furthermore, both the adhesive and the cork phases contributed to migration. Last, the more polar TEMPO-phthalate (μ, Table I) migrated more into 12% ethanol, despite its higher molecular weight.

Sorption Profile of the Simulant in the Cork

After immersion for 10 days at 40°C, corks were stored in a freezer to avoid evaporation. Samples were then cut off at different depths and weighed (by thermogravimetric analysis) to determine the diffusion profile of the simulant. The slope of this diffusion profile (Fig. 7) was small, which corresponded to weak migration profiles (Table II). A quick equilibration all over the cork could be envisaged.

Behavior of the Grafted AT Probe

AT, which grafted on the adhesive before cork processing, was a powerful tool used to study selectively the interaction between the adhesive and the simulant. The grafted probe could obviously neither migrate nor diffuse in the cork phase.

The grafted adhesive was allowed to polymerize as a film by exposure to atmospheric moisture. This film was then analyzed.

The spectrum of grafted adhesive–AT is shown in Figure 8. Recorded at room temperature, it appeared as a broad dissymmetric spectrum (hindered probe, T_{50G} = 135°C). When the temperature increased, a second signal became apparent (free probe, t_{fs} = 72°C).

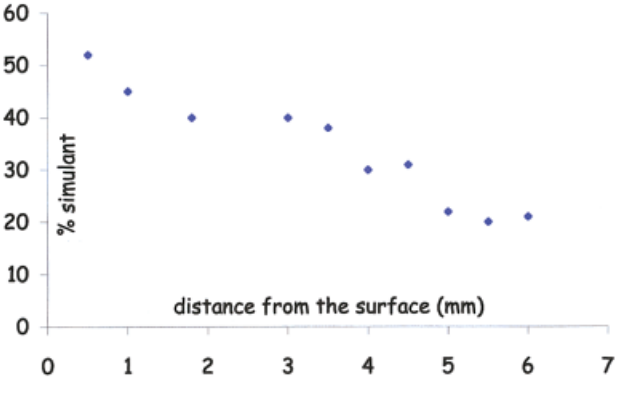

Figure 7 Diffusion profile of the simulant in the cork (10 days at 40°C).

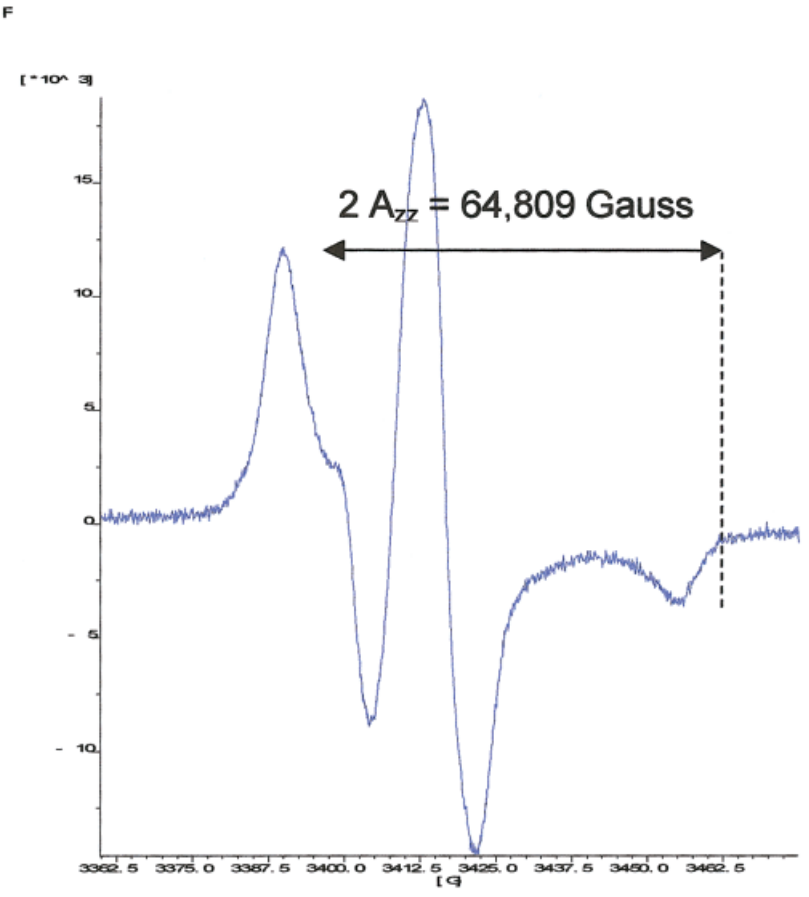

Figure 8 ESR spectrum of grafted adhesive–AT.

Several probe populations existed together in the adhesive. The main one, corresponding to T_{50G} = 135°C, corresponded to the grafted adhesive–AT. As grafting consumed a terminal N═C═O group, the grafted probe was necessarily located at a chain end. The second environment, corresponding to the t_{fs} could, in principle, be explained in several ways that are related to the presence of unreacted AT in the polymerized adhesive :

AT may have reacted with free toluene diisocyanide, producing a grafted monomer. The second environment could not be allocated to this molecule, as the mobility of NO° was linked to a rotation around the (aromatic ring)—N bond, which was similar in the grafted adhesive and in the grafted monomer.

Polyurethanes are known to display several transitions, which can also be observed by ESR.[14] With this particular material, this should have also been observed with the other probes, and this hypothesis could thus be ruled out.

Free AT was expected to form strong hydrogen bonds with the dioxyethylene units (OCH_2CH_2O) of the polymer (Fig. 2). Such hydrogen bonds are known to restrict the mobility of hydrogen-donating probes. This explains nicely why the free AT had a higher transition temperature than DOXYL-4,4 and TEMPO-phthalate, which are not hydrogen donors (Table III). This behavior of free AT, the formation of hydrogen bonds with additives or with polymers in polymeric materials, has already been observed.[15,16]

Table III Mobility of the Probes in the Adhesive

Probe	T_{50G} (°C)
TEMPO-phthalate	17
DOXYL-4,4	< 17
DOXYL-9,9	< 17
AT	72[a]

[a] t_{fs} observed in the grafted adhesive.

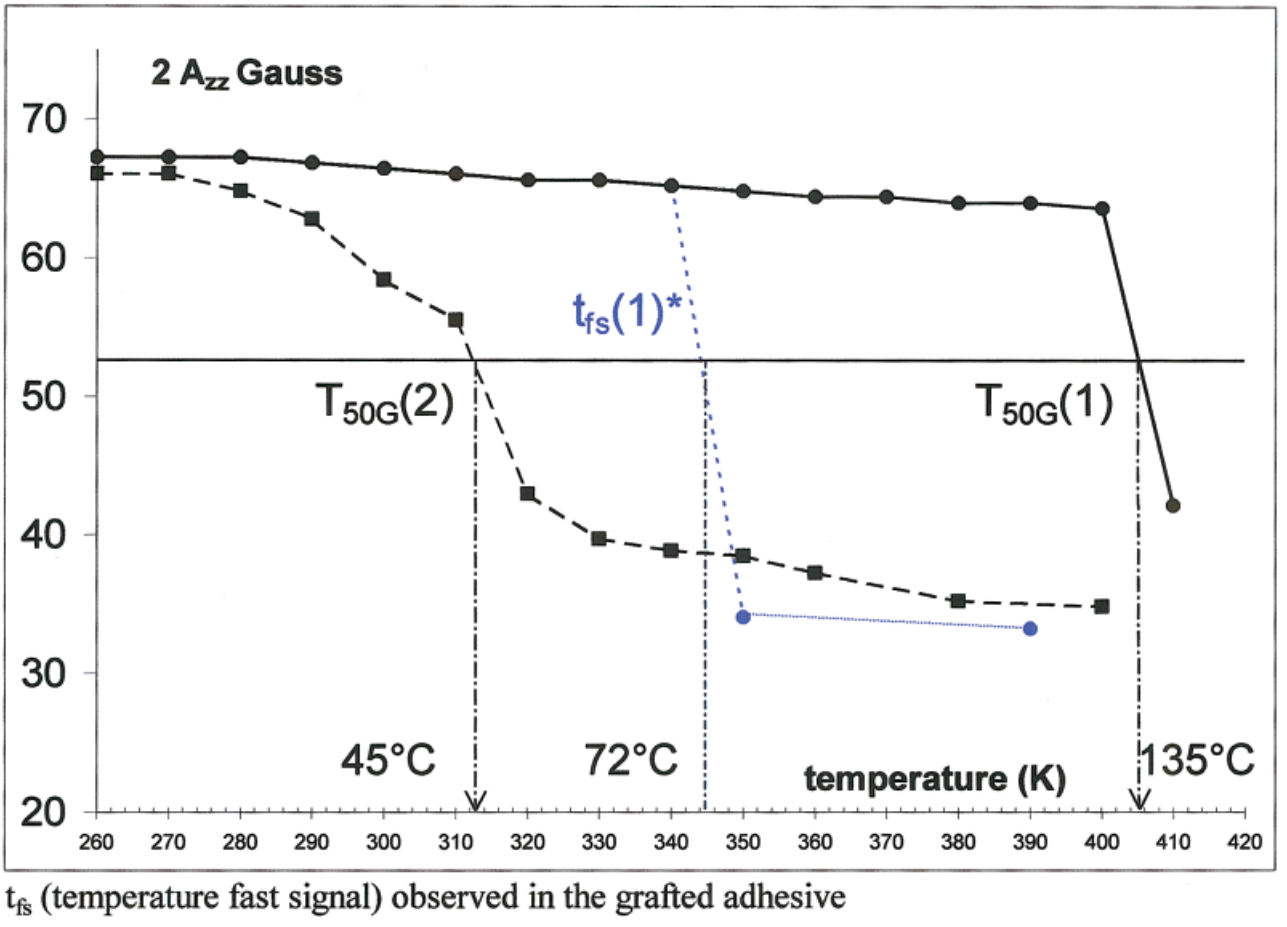

Figure 9 Mobility (T_{50G}) of the probe AT in the adhesive (1) before and (2) after contact with ethanol 12%.

After contact with the simulant (10 days at 40°C), T_{50G} strongly decreased (T_{50G} = 45°C; Fig. 9). This result reveals that after contact, the structure of the adhesive was altered and that AT could reorient more easily. This increased mobility was assigned to the penetration of the simulant into the adhesive phase, which modified the environment of the probe:[9,10] 12% ethanol plasticized the adhesive, increased the mobility of the probe, and probably also enhanced its migration.

This plasticization was confirmed by DSC, which showed a decrease of the lower transition from −52 to −56°C after contact with the simulant. The shift in transition temperatures was much less important with ESR, which must be related to the fact that DSC analyzes the bulk, whereas ESR is a local observation around chain ends.

These results suggest that migration occurred through the plasticization of the adhesive by the wine simulant, associated to a diffusion of small molecules into the cork phase.

CONCLUSIONS

Probes incorporated into the adhesive partition in the cork phase during processing at high temperature. Consequently, the whole finished cork must be considered as a reservoir of potential migrants.

Partition ratios (cork/adhesive) tended to follow the polarity of the probes.

The strong penetration of the simulant (12% ethanol) in the whole structure of the finished cork greatly facilitated migration of probes and further organic molecules.

The authors thank the Comité Interprofessionnel des Vins de Champagne, the Syndicat des Bouchonniers en

Champagne, Ato Bostik-Findley, for a grant to T. Six and Europol' Agro for financial support of the project.

REFERENCES

1. Six, T. Ph.D. Thesis, Université des Sciences de Reims, November 17, 2000.
2. Council of Europe. Resolution on Cork Intended to Come into Contact with Foodstuff; 2000.
3. Grob, K.; Spinner, C.; Brunner, M.; Etter, R. Food Additives Contam 1999, 16, 12.
4. Hadcock, L. H. Plast Rubber Appl 1984, 4, 53.
5. Metois, P.; Scholler, D.; Bouquant, J.; Feigenbaum, A. Food Additives Contam 1998, 15, 1.
6. Riquet, A. M.; Wolff, N.; Laoubi, S.; Vergnaud, J. M.; Feigenbaum, A. Food Additives Contam 1998, 15, 6.
7. Araujo, C. D.; MacKay, A. L.; Hailey, J. R. T.; Whitall. K. P. Wood Sci Technol 1992, 26, 101.
8. Figge, K. Food Cosmet Toxicol 1972, 10, 815.
9. Riquet, A. M.; Sandray, V.; Akermann, O.; Feigenbaum, A. Sci Aliments 1991, 11, 2.
10. Riquet, A. M.; Hamdani, M.; Feigenbaum, A. J Polym Eng 1995/1996, 15, 1.
11. Berliner, L. Spin Labelling. Theory and Applications; Academic: London, 1976; Volume 1.
12. Cottier, S.; Riquet, A.M.; Feigenbaum, A.; Mortreuil, P. J Appl Polym Sci 1996, 62, 2219.
13. Hamdani, M., personal communication.
14. Chen, W. P.; Schlick, S. Polymer 1990, 31, 308.
15. Hamada, K.; Iijima, T.; McGregor, R. Polym J 1987, 9, 709.
16. Riquet, A. M.; Wolff, N.; Feigenbaum, A. Eur Polym J 1992, 24, 4.